

Amendment to the Claims:

Please amend the claims as follows:

Please cancel claims 8, 9, 12, 14 to 20, 26, 28 to 30, 32, 33, 35, 39, 43, 44, 46, 48 to 105, 107 to 125, 127, 129 to 150, 152 to 166, 168 to 196 and 198 to 258, without prejudice or disclaimer.

This listing of claims will replace all prior versions, and listing, of claims in the application:

Listing of Claims:

Claim 1 (currently amended): An isolated, synthetic or recombinant nucleic acid comprising

(a) a nucleic acid sequence having at least ~~[[50%]]~~ 90% sequence identity to ~~SEQ ID NO:1, SEQ ID NO:3, SEQ ID NO:5, SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23 or SEQ ID NO:25,~~ over a region of at least ~~[[about 100]]~~ 1650 residues, wherein the nucleic acid encodes ~~at least one~~ a polypeptide having a laccase activity, or

(b) a nucleic acid sequence completely complementary to (a)  
~~and the sequence identities are determined by analysis with a sequence comparison algorithm or by a visual inspection.~~

Claim 2 (currently amended): The isolated, synthetic or recombinant nucleic acid of claim 1, wherein the sequence identity is at least 95% ~~about 51%, 52%, 53%, 54%, 55%, 56%, 57%, 58%, 59%, 60%, 61%, 62%, 63% or 64%.~~

Claim 3 (currently amended): The isolated, synthetic or recombinant nucleic acid of claim 1, wherein the sequence identity is at least ~~about 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or more~~ sequence identity to ~~SEQ ID NO:1, SEQ ID NO:3, SEQ ID NO:5, SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23 or SEQ ID NO:25.~~

Claim 4 (currently amended): The isolated, synthetic or recombinant nucleic acid of claim 1, wherein the percentage sequence identity is over a region of at least ~~about 50, 75, 100, 150, 200, 250, 300, 350, 400, 450, 500, 550, 600, 650, 700, 750, 800, 850, 900, 950, 1000, 1050, 1100, 1150~~ 1700 bases ~~or more residues, or the full length of a gene or a transcript.~~

Claim 5 (currently amended): The isolated, synthetic or recombinant nucleic acid of claim 1, wherein the nucleic acid sequence comprises the ~~the~~ [[a]] ~~sequence of~~ as set forth in SEQ ID NO:1, SEQ ID NO:3, SEQ ID NO:5, SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23 or SEQ ID NO:25.

Claim 6 (currently amended): The isolated, synthetic or recombinant nucleic acid of claim 1, wherein the nucleic acid comprises a sequence that encodes at least 550 contiguous amino acids of a polypeptide ~~having comprising the~~ comprising the [[a]] ~~sequence of~~ as set forth in SEQ ID NO:2, SEQ ID NO:4, SEQ ID NO:6, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:18, SEQ ID NO:20, SEQ ID NO:22, SEQ ID NO:24, SEQ ID NO:26.

Claim 7 (currently amended): The isolated, synthetic or recombinant nucleic acid of claim 1, wherein the sequence identities are determined by analysis with a sequence comparison algorithm comprising ~~comprising~~ [[is]] a BLAST version 2.2.2 algorithm where a filtering setting is set to blastall -p blastp -d "nr pataa" -F F, and all other options are set to default.

Claims 8 to 9 (canceled)

Claim 10 (currently amended): The isolated, synthetic or recombinant nucleic acid of claim 1, wherein the laccase activity comprises catalyzing the oxidation ~~of 1-~~ hydroxybenzotriazole (HBT), N-benzoyl-N-phenylhydroxylamine (BPHA), N-hydroxyphthalimide, 3-hydroxy-1,2,3-benzotriazin-4-one, promazine, 1,8-dihydroxy-4,5-dinitroanthraquinone, phenoxazine, anthraquinone, 2-hydroxy-1,4-naphthoquinone, phenothiazine, syringaldazine, anthrone, anthracene, anthrarufin, anthrarobin, 2,2'-azino-bis(3-ethylbenzthiazoline-6-sulfonic acid) (ABTS), dimethoxyphenol (DMP), ferulic acid, catechin,

~~epicatechin, homovanillic acid (HMA), 2,3-dihydroxybenzoic acid (2,3-DHB), 2,2,6,6-tetramethylpiperidin-1-yloxy (TEMPO), dimethoxyphenol or dihydroxyfumaric acid (DHF).~~

Claim 11 (currently amended): The isolated, synthetic or recombinant nucleic acid of claim 1, wherein the laccase activity comprises a peroxidase activity ~~the depolymerization of a cellulose or a cellulose derivative or a hemicellulose.~~

Claim 12 (canceled)

Claim 13 (currently amended): The isolated, synthetic or recombinant nucleic acid of claim 1, wherein the laccase activity ~~comprises production of a norketone from a~~ comprises oxidation of valencene.

Claims 14 to 20 (canceled)

Claim 21 (currently amended): The isolated, synthetic or recombinant nucleic acid of claim ~~1, 20~~, wherein the laccase activity comprises oxidation of a polyphenol, a methoxy-substituted monophenol, an aromatic amine, or an oxidizable aromatic compound.

Claim 22 (currently amended): The isolated, synthetic or recombinant nucleic acid of claim 1, wherein the polypeptide retains a laccase activity after exposure to a temperature range of 55°C and 75°C.

Claim 23 (currently amended): The isolated, synthetic or recombinant nucleic acid of claim ~~1, 22~~, wherein the polypeptide retains a laccase activity under conditions comprising a temperature range of ~~between about 37°C to about 95°C, or between about 55°C to about 85°C, or between about 70°C to about 75°C, or between about 70°C to about 95°C, or between about 90°C to about 95°C, or, the polypeptide retains a laccase activity after exposure to a temperature in the range from greater than 37°C to about 95°C, from greater than 55°C to about 85°C, or between about 70°C to about 75°C, or from greater than 90°C to about 95°C.~~

Claim 24 (currently amended): An isolated, synthetic or recombinant nucleic acid, wherein the nucleic acid comprises

(a) a sequence at least 1650 bases in length that hybridizes under stringent conditions to a nucleic acid comprising ~~SEQ ID NO:1, SEQ ID NO:3, SEQ ID NO:5, SEQ ID NO:7, SEQ ID~~

~~NO:9, SEQ ID NO:11, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23 or SEQ ID NO:25, wherein the nucleic acid encodes a polypeptide having a laccase activity, and wherein the stringent conditions include a wash step comprising a wash in 0.2X SSC at a temperature of about 65°C for about 15 minutes, or~~

(b) a nucleic acid sequence completely complementary to (a).

Claim 25 (currently amended): The isolated, synthetic or recombinant nucleic acid of claim 24, wherein the sequence nucleic acid is at least 1700 bases ~~about 50, 75, 100, 150, 200, 300, 400, 500, 600, 700, 800, 900, 1000 or more residues in length or the full length of the gene or transcript.~~

Claim 26 (canceled)

Claim 27 (currently amended): A nucleic acid probe for identifying a nucleic acid encoding a polypeptide with a laccase activity, wherein the probe comprises at least ~~40~~ 60 to 150 consecutive bases of a nucleic acid sequence having at least 90% sequence identity to a subsequence of a sequence comprising SEQ ID NO:1, SEQ ID NO:3, SEQ ID NO:5, SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23 or SEQ ID NO:25, wherein the probe identifies the nucleic acid by binding or hybridization.

Claims 28 to 30 (canceled)

31. (currently amended) An amplification primer pair for amplifying a nucleic acid encoding a polypeptide having a laccase activity, wherein the primer pair is capable of amplifying a nucleic acid comprising the [[a]] sequence as set forth in of claim 1 or 24, or a subsequence thereof, wherein the first member comprises ~~about~~ at least the first (the 5') 12 bases of SEQ ID NO:23, and the second member comprises at least the first (the 5') 12 bases of the complementary strand of SEQ ID NO:23.

Claims 32 to 33 (canceled)

34. (currently amended) A laccase-encoding nucleic acid generated by amplification of a polynucleotide using an amplification primer pair ~~as set forth in of claim 31~~ 33.

Claim 35 (canceled)

36. (currently amended) The laccase-encoding nucleic acid of claim 34, wherein the nucleic acid is generated by amplification of a gene library.

37. (currently amended) The laccase-encoding nucleic acid of claim 36, ~~34~~, wherein the gene library is an environmental library.

38. (currently amended) An isolated, synthetic or recombinant laccase encoded by the ~~[[a]]~~ laccase-encoding nucleic acid ~~as set forth in~~ of claim 1 ~~[[34]]~~.

Claim 39 (canceled)

40. (currently amended) An expression cassette comprising ~~a~~ the nucleic acid ~~comprising a sequence as set forth in~~ of claim ~~1 or claim 24~~.

41. (currently amended) A vector comprising ~~a~~ the nucleic acid ~~comprising a sequence as set forth in~~ of claim ~~1 or claim 24~~.

42. (currently amended) A cloning vehicle comprising ~~a~~ the nucleic acid ~~comprising a sequence as set forth in~~ of claim ~~1 or claim 24~~, wherein the cloning vehicle comprises ~~a viral vector, a plasmid, a phage, a phagemid, a cosmid, a fosmid, a bacteriophage or an artificial chromosome.~~

Claims 43 to 44 (canceled)

45. (currently amended) A transformed cell comprising ~~a~~ the nucleic acid ~~comprising a sequence as set forth in~~ of claim ~~1 or claim 24~~.

Claim 46 (canceled)

47. (currently amended) The transformed cell of claim 45, ~~40~~, wherein the cell is a ~~bacterial cell, a mammalian cell, a fungal cell, a yeast cell, an insect cell or a plant cell.~~

Claims 48 to 105 (canceled)

106. (currently amended) A method ~~for~~ of producing a recombinant polypeptide having a laccase activity, comprising the steps of:

- (a) transforming a host cell with ~~providing~~ a nucleic acid operably linked to a promoter, wherein the nucleic acid comprises the [[a]] sequence as set forth in of claim 1 or claim 24; and
  - (b) expressing the nucleic acid of step (a) under conditions that allow expression of the polypeptide,
- thereby producing the [[a]] recombinant polypeptide.

Claims 107 to 125 (canceled)

126. (currently amended) A method for isolating or recovering a nucleic acid encoding a polypeptide with a laccase activity from an environmental sample, comprising the steps of:

- (a) providing ~~[[an]] the~~ amplification primer ~~sequence pair as set forth in of claim 31 or claim 33~~;
  - (b) isolating a nucleic acid from the environmental sample or treating the environmental sample ~~such~~ so that nucleic acid in the sample is accessible for hybridization to the amplification primer pair; and,
  - (c) combining the nucleic acid of step (b) with the amplification primer pair of step (a) and amplifying nucleic acid from the environmental sample,
- thereby isolating or recovering a nucleic acid encoding a polypeptide with a laccase activity from an environmental sample.

Claim 127 (canceled)

128. (currently amended) A method for isolating or recovering a nucleic acid encoding a polypeptide with a laccase activity from an environmental sample, comprising the steps of:

- (a) providing ~~a polynucleotide the~~ probe of claim 27 ~~comprising a sequence as set forth in claim 1 or claim 24, or a subsequence thereof~~;
- (b) isolating a nucleic acid from the environmental sample or treating the environmental sample ~~such~~ so that the nucleic acid ~~in the sample~~ is accessible for hybridization to ~~a polynucleotide the~~ probe of step (a);

- (c) combining the isolated nucleic acid or the treated environmental sample of step (b) with the polynucleotide probe of step (a); and
- (d) isolating a nucleic acid that specifically hybridizes with the polynucleotide probe; of step (a);

thereby isolating or recovering a nucleic acid encoding a polypeptide with a laccase activity from an environmental sample.

Claims 129 to 150 (canceled)

151. (currently amended) A method for ~~modifying~~ oxidizing an aromatic amine a small molecule, comprising the following steps:

- (a) providing ~~a laccase enzyme, wherein the enzyme comprises a polypeptide as set forth in claim 64, or a polypeptide encoded by [[a]] the nucleic acid comprising a nucleic acid sequence as set forth in of claim 1 or claim 24;~~
- (b) providing an aromatic amine a small molecule; and
- (c) reacting the polypeptide enzyme of step (a) with the aromatic amine small molecule of step (b) under conditions that facilitate ~~an enzymatic reaction~~ catalyzed by the laccase activity of the polypeptide; laccase enzyme,

thereby modifying oxidizing the aromatic amine a small molecule by a laccase enzymatic reaction.

Claims 152 to 166 (canceled)

167. (currently amended) The [[A]] method of increasing thermotolerance or thermostability of a laccase polypeptide comprising, claim 106, wherein step (b) comprises glycosylating the polypeptide a laccase, wherein the polypeptide comprises at least thirty contiguous amino acids of a polypeptide as set forth in claim 60, or a polypeptide encoded by a nucleic acid as set forth in claim 1 or claim 24, thereby increasing the thermotolerance or thermostability of the laccase.

Claims 168 to 196 (canceled)

197. (currently amended) ~~The An~~ isolated, synthetic or recombinant nucleic acid of claim 267, further comprising a sequence encoding a polypeptide having a lacease activity and a signal sequence, wherein the nucleic acid comprises a sequence as set forth in claim 1.

Claims 198 to 258 (canceled)

259. (currently amended) The method of [[as]] claim 106 [[107]], wherein the host cell is ~~selected from the group consisting of a plant cell, a bacterial cell, a yeast cell, an insect cell, or and an animal cell.~~

260. (currently amended) The method of claim 259, wherein the host is selected from the group consisting of a *Schizosaccharomyces* sp., *Saccharomyces* sp., and *Pichia* sp., ~~*Escherichia* sp., *Streptomyces* sp., *Bacillus* sp. and *Lactobacillus* sp.~~

261. (currently amended) The method of claim 260, wherein the ~~organism~~ host is ~~S.~~ *Schizosaccharomyces pombe*.

262. (currently amended) The method of claim 260, wherein the ~~organism~~ host is ~~S.~~ *Saccharomyces cerevisiae*.

263. (currently amended) The method of claim 260, wherein the ~~organism~~ host is ~~P.~~ *Pichia pastoris*.

264. (currently amended) The method of claim 106, ~~260~~, wherein the host cell ~~organism~~ is *E. coli*.

265. (currently amended) The method of claim 106, ~~260~~, wherein the host cell ~~organism~~ is *Bacillus cereus*.

266. (new) The nucleic acid of claim 6, wherein the nucleic acid comprises a sequence encoding the polypeptide sequence of SEQ ID NO:24.



267. (new) An isolated, synthetic, or recombinant nucleic acid comprising a sequence having at least 90% identity to at least 1700 bases, over the region of nucleotide residue 60 to 1767 of SEQ ID NO:23, wherein the nucleic acid encodes a polypeptide having a laccase activity.

268. (new) The nucleic acid of claim 267, wherein the identity is at least 95% to the region of nucleotide residue 60 to 1767 of SEQ ID NO:23.

269. (new) The nucleic acid of claim 267, wherein the identity is 100% for at least 1700 consecutive nucleotide residues of SEQ ID NO:23.

270. (new) The method of claim 151, wherein the aromatic amine is 2,2'-azino-bis(3-ethylbenzthiazoline-6-sulfonic acid) (ABTS).

271. (new) A method for oxidizing valencene, comprising the following steps:

- (a) providing a polypeptide encoded by the nucleic acid of claim 1;
- (b) providing valencene; and
- (c) reacting the polypeptide of step (a) with the valencene under conditions that facilitate the laccase activity of the polypeptide;

thereby oxidizing the valencene.

272. (new) The nucleic acid of claim 267, wherein the sequence identities are determined by analysis with a sequence comparison algorithm comprising a BLAST version 2.2.2 algorithm where a filtering setting is set to blastall -p blastp -d "nr pataa" -F F, and all other options are set to default.